1: *Gut* 1999 Mar;44(3):366-71

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Adenovirus mediated p53 tumour suppressor gene therapy for human gastric cancer cells in vitro and in vivo.

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BACKGROUND/AIMS: Gastric cancer is one of the most prevalent forms of cancer in East Asia. Point mutation of the p53 gene has been reported in more than 60% of cases of gastric cancer and can lead to genetic instability and uncontrolled cell proliferation. The purpose of this investigation was to evaluate the potential of p53 gene therapy for gastric cancer. METHODS: The responses of human gastric cancer cell lines, MKN1, MKN7, MKN28, MKN45, and TMK-1, to recombinant adenoviruses encoding wild type p53 (AdCAp53) were analysed in vitro. The efficacy of the AdCAp53 treatment for MKN1 and MKN45 subcutaneous tumours in nude mice was assessed in vivo. RESULTS: p53-specific growth inhibition was observed in vitro in two of four gastric cancer cell lines with mutated p53, but not in the wild type p53 cell line. The mechanism of the killing of gastric cancer cells by AdCAp53 was found, by flow cytometric analysis and detection of DNA fragmentation, to be apoptosis. In vivo studies showed that the growth of subcutaneous tumours of p53 mutant MKN1 cells was significantly inhibited by direct injection of AdCAp53, but no significant growth inhibition was detected in the growth of p53 wild type MKN45 tumours. CONCLUSIONS: Adenovirus mediated reintroduction of wild type p53 is a potential clinical utility in gene therapy for gastric cancers.

PMID: 10026322, UI: 99152130

1: *Cancer Res* 1997 Oct 1;57(19):4279-84 Related Articles, Books LinkOut

In vivo selective gene expression and therapy mediated by adenoviral vectors for human carcinoembryonic antigen-producing gastric carcinoma.

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Previously, we reported that adenoviral vectors carrying the carcinoembryonic antigen (CEA) promoter sequences to direct the Echerichia coli beta-galactosidase gene (AdCEA-lacZ) or cytosine deaminase (CD) gene (AdCEA-CD) confer selective gene expression on a CEA-positive gastric cancer cell line (MKN45) in vitro. Here, adenovirus-mediated tumor-specific gene therapy for CEA-positive gastric carcinoma in vivo was investigated. Using an animal model with i.p. disseminated MKN45 tumors, adenovirus-mediated tumor-specific transgene expression and therapeutic efficacy were analyzed. After an i.p. injection of AdCEA-lacZ, beta-galactosidase activity was confined to tumor xenografts. Moreover, CD mRNA was expressed exclusively in MKN45 tumor xenografts after infection with AdCEA-CD, despite the fact that an adenovirus-mediated transfer of CD DNA was detected in all tissues tested. In contrast, CD mRNA was detected not only in tumor xenografts but also in other organs of mice infected with AdCA-CD, in which CD gene expression is governed by an ubiquitous promoter. Suppression of tumor growth and prolongation of survival were noted in tumor-bearing mice treated with AdCEA-CD and 5-fluorocytosine (5FC) without observable adverse effects. In contrast, significant hepatic toxicity was noted in animals treated with AdCA-CD. These results reveal that the CEA promoter restricts CD gene expression to CEA-positive tumor cells in the adenoviral context in vivo, along with the beneficial therapeutic effects of 5FC treatment, suggesting the i.p. AdCEA-CD/5FC system may provide a novel approach to treatment of i.p. disseminated gastric cancer.

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1: *Cancer Res* 1997 Apr 1;57(7):1335-43

The anti-human tumor effect and generation of human cytotoxic T cells in SCID mice given human peripheral blood lymphocytes by the in vivo transfer of the Interleukin-6 gene using adenovirus vector.

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Interleukin-6 (IL-6) was found to function as a late-acting killer helper factor in the differentiation of CTLs. In the model of tumor-bearing mice, the systemic administration of recombinant IL-6 was found to mediate the antitumor effect on the immunogenic murine tumors via the in vivo induction of murine CTLs but not on the poorly immunogenic murine tumors in our previous study. However, an in vivo experimental model capable of analyzing the anti-human tumor effect via the in vivo induction of human CTLs has not yet been established. Therefore, in the present study, severe combined immunodeficient mice were given human peripheral blood lymphocytes (SCID-PBL/hu), and thereafter human tumor cells were administered i.p. into these SCID-PBL/hu mice as a model of human patients with cancer. When these SCID-PBL/hu mice bearing allogeneic human CESS B blastoid tumor cells were treated in vivo with recombinant adenovirus vector expressing IL-6 cDNA, both the induction of CD8+ human CTLs against CESS cells in the spleen cells and peritoneal exudate cells and a prolongation in the survival of these mice were observed. Furthermore, SCID-PBL/hu mice were given peripheral blood lymphocytes from patients with cancer (gastric or rectal cancers) and autologous human tumor cells. The in vivo administration of recombinant adenovirus vector expressing IL-6 cDNA induced CD8+ human CTLs specific for autologous human tumor cells from human precursor T cells. The in vivo injection of the IL-6 gene also inhibited growth and metastasis in autologous human cancers. Based on the above findings, the experimental model using SCID-PBL/hu mice and the IL-6 gene delivered in vivo by an adenovirus vector might therefore provide a new strategy capable of analyzing an anti-human tumor effect and the in vivo induction of human CTLs by cytokine gene therapy without using the human body.

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1: *Biochem Biophys Res Commun* 1997 Feb 24;231 (3):775-9

IDEAL

Adenovirus-mediated gene therapy of gastric carcinoma using cancer-specific gene expression in vivo.

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The carcinoembryonic antigen (CEA) is a glycoprotein which overexpressed in the majority of human gastric cancers. We demonstrated that recombinant adenoviral vector (AdCEAtk), containing the CEA promoter, could transfer the herpes simplex virus thymidine kinase (HSVtk) gene into CEA-producing gastric cancer cells to confer sensitivity to ganciclovir (GCV) in vivo. In an ex vivo experiment, the tumor growth was inhibited after GCV treatment when the tumor contained more than 20% of AdCEAtk infected cells, indicating an efficient bystander killing effect. With intra-tumoral injection of AdCEAtk, the HSVtk were selectively expressed in approximately 30% of CEA producing cancer cells. By AdCEAtk injection and GCV administration, the growth of tumors was significantly inhibited by 20% as compared to untreated tumors. It is hoped that these results provide a strategy of tumor specific gene transfer for CEA producing gastric cancers.

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1: *Hum Gene Ther* 1996 Mar 20;7(5):589-93

In vivo gene transfer and expression in rat stomach by submucosal injection of plasmid DNA.

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Gastrointestinal nonepithelial tissue is a useful target for in vivo gene transfer. The aim of this study was to investigate whether gene transfer into this organ could be achieved by submucosal injection of plasmid DNA. Plasmid DNA carrying either the firefly luciferase or Escherichia coli LacZ reporter gene was injected directly into the gastric submucosa of adult rats. Gene expression was characterized by quantitative luciferase assay and qualitative in situ beta-galactosidase (beta-Gal) staining. Luciferase activity was detected as early as 1 day after injection, increased markedly at 2 days, and then decreased. Some of the rats showed detectable levels of luciferase expression at 14 and 21 days postinjection. Histochemical staining for beta-Gal demonstrated that expression of the recombinant genes was localized in smooth muscle cells of the muscularis mucosae and the muscular layer and mesenchymal cells in the lamina propria. Our results indicate that gene transfer into the gastrointestinal tract can be achieved by simple needle insertion of naked plasmid DNA into the submucosa.

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